Increased photosynthetic performance of *Brassica napus* contributes to enhanced Cd phytomanagement under warmer climate despite drought stress

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The world growing population with the increased anthropogenic activities such as mining, smelting, mismanagement of metal waste, disposal of manure, wastewater and sewage sludge, indiscriminate use of phosphate fertilizers and pesticides has resulted in widespread contamination of cadmium (Cd) and other heavy metals (HMs) in the environment (Dutta et al., 2021; Haider et al., 2021; Yan et al., 2020). In addition, a steady rise in atmospheric CO_2 concentration has driven a dangerous rise in global land surface temperature, termed global warming, which is driving an alarming increase in the frequency and intensity of different abiotic stresses, such as drought (Rivero et al., 2021). Consequently, at this time, key ecosystems and ecological processes are simultaneously experiencing pollution-related and climate-related stresses. Because HMs are non-biodegradable, and immobilization may only reduce their bioavailability temporarily, as the soil environment may change and they may become mobile again for plants to take up, the most appropriate technique for removing Cd from contaminated soils is to remove Cd from the soil (Yan et al., 2020). Phytoextraction is such a plant-based technique where plants are used to translocate HMs from soil into the aboveground harvestable biomass (Khalid et al., 2017). Unlike conventional physical and chemical methods, phytoextraction has been considered to be among the safest, cleanest, cost–effective, sustainable, and least disruptive options for treating sites contaminated with recalcitrant pollutants like HMs (Diarra et al., 2021). *Brassica napus* is an emerging biofuel crop that can be used to produce biofuel when grown in degraded soil and has many features suitable for the phytomanagement of Cd-contaminated soils (Rizwan et al., 2018). However, the ongoing climate change with the increasing drought periods is bound to have an impact on phytoextraction performance, yet there remains little research on this. Therefore, the aim of this study was to investigate Cd phytoextraction efficiency by *B. napus* under drought stress in current and future warmer climate conditions.

The experiment was carried out in a completely random design with three replications at Vytautas Magnus University (Lithuania) growth chambers with a volume of 10 m^3 each in a controlled environment for a period of 64 days. *B. napus* plants were sown in 3-liter vegetation pots filled with field topsoil, agroperlite and sand in a 5:3:2 volume ratio under the weight of 2.5 kg spiked with different concentrations (0, 1, 10, 50, and 100 mg kg^{-1}) of cadmium chloride ($CdCl_2 \times 2.5\ H_2O$). Then half of the pots were placed in a growth chamber under the current climate conditions (21/14 °C day/night, 400 μmol mol^{-1} CO_2 and 55-60/65-70% relative air humidity (RH)), while another half of the pots under future warmer climate conditions (25/18 °C, 800 μmol mol^{-1} CO_2 and 45-50/55-60% RH). Drought stress to half of the pots in each chamber was applied on the 46th day after sowing by withholding watering until the volumetric soil water content (SWC) dropped to an average of 5%. Then, drought-stressed plants were kept at 5% of SWC till the end of the experiment, while control plants were maintained at 30% of SWC. Cd concentration in mineralized samples (Milestone ETHOS One, Italy) was determined by the inductively coupled plasma–optical emission spectrometry method (Optima 8000, PerkinElmer, USA). Cd removal efficiency, defined here as Cd accumulation, was determined by the harvestable plant biomass (shoot dry weight, DW) multiplied by the concentration of Cd contained within this biomass. Photosynthetic performance of *B. napus* was evaluated using gas exchange parameters, measured with LI-6400 infrared gas-exchange system (Li-6400, LI-COR Inc., NE, USA).

The obtained results showed that, under both water regimes, in higher Cd treatments (50 and 100 mg kg^{-1}), the future warmer climate conditions resulted in lower Cd concentration in *B. napus* shoot dry weight when compared to those grown under current climate conditions ($p < 0.05$) (Fig. 1C). However, under both water regimes, warmer climate conditions allowed to significantly increase the photosynthetic rate of *B. napus* (Fig. 1A), which translated into considerably higher aboveground biomass production in all Cd treatments under well-watered conditions and in lower Cd treatments (1 and 10 mg kg^{-1}) under drought conditions (Fig. 1B). The latter fact led to highly increased (up to 65-76%, $p < 0.05$) Cd accumulation in the aboveground biomass of well-watered *B. napus* plants grown under future warmer climate conditions in higher Cd treatments (50 and 100 mg kg^{-1}) (Fig 1D). Drought diminished the positive warmer climate impact on Cd phytoextraction by *B. napus*. However, although drought-stressed plants of *B. napus* grown under warmer climate conditions did not extract significantly higher Cd content, compared to those under current climate conditions, the accumulated Cd content in their shoot dry weight also did not differ significantly from well-watered ones grown under current climate conditions (Fig. 1D). Taking all into account, the findings of this study revealed that *B. napus* has the potential to be more efficient for Cd phytomanagement purposes at higher Cd levels in soil under future warmer climate conditions, even in the presence of drought stress.

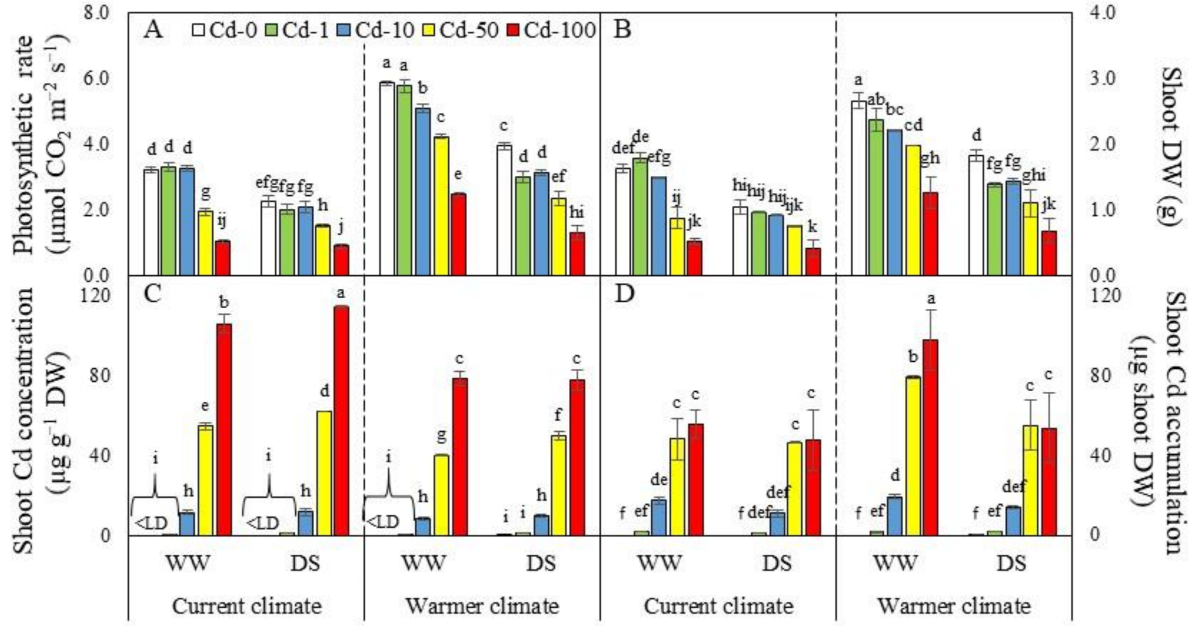

Figure 1. (A) Photosynthetic rate, (B) shoot dry weight (DW), (C) shoot Cd concentration, and (D) shoot Cd accumulation of well-watered (WW) and drought-stressed (DS) *B. napus* plants grown under different Cd concentrations (0, 1, 10, 50, and 100 mg Cd kg^{-1}) in current (21/14 °C, 400 µmol mol^{-1} CO_2 and 55-60/65-70% RH) and warmer climate (25/18 °C, 800 µmol mol^{-1} CO_2 and 45-50/55-60% RH) conditions. Data are means ± SE (for photosynthetic rate n = 6-9, for other parameters n = 3); different letters outside the bars indicate significant differences among the treatments ($p < 0.05$, Fisher's LSD). <LD – below detection limit

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